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Intraspecific variation in *Anaea ryphea* Cramer and *Anaea eurypyle* C. and R. Felder (Nymphalidae).

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Abstract. *Anaea ryphea* and *Anaea eurypyle* are butterflies with wide intraspecific variation, and similar ranges, Mexico to Southern Brazil. The two characters used to separate the two taxa are continually variable, i.e., there are no well defined states for them. The frequency distributions for each of these two characters is unimodal when individuals of both species are combined. Specimens identified as *A. eurypyle* have wing patterns on one extreme of the distributions, and at most localities the majority of individuals have the *A. ryphea* phenotype, which includes the mode of both frequency distributions. Results of my research so far suggest that these two taxa may in fact represent one highly variable species, or if they are distinct biological species, they have considerable character overlap.

Introduction

Taxonomic knowledge of the genus *Anaea* Hübner (Nymphalidae) is based mainly on a treatment by Comstock (1961). Many of the species originally described there have since been split into other genera, including taxa treated by Comstock as subgenera (Descimon 1986) and other taxa described later (Rydon 1971). Although *Anaea* is typical of neotropical areas and has widespread distribution (Comstock 1961; De Vries 1987), the species comprising this genus have received little or no study. For example, reports of the larval food plant and aspects of the biology and population ecology of *Anaea ryphea* Cramer (Caldas 1991, 1994; 1995a,b) were not published until after D'Abrera's volume V, Nymphalidae and Satyridae, Butterflies of the Neotropical Region (1988). In this same volume, D'Abrera describes the chaotic taxonomic situation in *Anaea*, and follows Rydon (1971) in maintaining *Memphis* and *Fountainea* as genera separate from *Anaea*.

Anaea ryphea is found from Mexico to Southern Brazil. Males vary greatly in wing color, pattern, and shape, and much of this variation was mentioned by Comstock (1961). I have been studying ecological aspects of *Anaea ryphea* populations since 1988. Data from Brazil (Caldas 1994), Panama (Caldas, unpublished data), and the literature (Muyshondt 1974) show that some variable characteristics of this taxon overlap with those of *Anaea eurypyle* C. and R. Felder. The larval stages of the two are virtually identical (Caldas 1994); they both use *Croton* sp. (Euphorbiaceae) as the larval food plant (DeVries 1987; Caldas 1991), and the morphological differences used to separate them are found mainly in male adults. Although they have the same geographical range, I have never (in 4 years in southern Brazil and one year in Panama) seen, collected, or reared one individual that I would identify as

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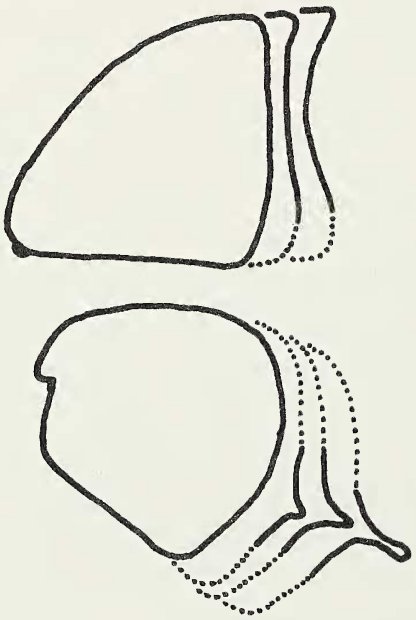
A. eurypyle. This situation led me to analyze museum specimens and search the literature in an attempt to discover characters diagnostic for these seemingly indistinguishable taxa.

I may add that, although intraspecific variation is fairly common in the Lepidoptera (Owen 1971; Vane-Wright & Ackery 1984) and is extremely important to the discussion of species concepts and updating of area checklists (Collins 1991), there are few studies on this subject in the literature (see Burns 1984 and 1992).

Materials and Methods

In an attempt to discover characters to distinguish *A. ryphea* and *A. eurypyle*, I examined specimens in the National Museum of Natural History (USNM), Smithsonian Institution, Washington, DC, and the American Museum of Natural History (AMNH), New York, where the original specimens used by Comstock (1961) are deposited. A total of 499 males from localities covering almost the entire range of distribution of the two species (both with two subspecies) were analyzed with respect to the four main variable external characters described by Comstock (1961): 1) Elongation ("production" of Comstock) of vein M3 of the hind wings to form a "tail" (presence/absence/length; Fig.1); 2) degree of acuteness of the forewing apex (straight to acute; Fig.1); 3) medial ("mesial" of Comstock) line on underside of the hind wings (straight to irregular; Fig.2); 4) white and black markings on the undersides of both wings (presence/absence). Individuals that had either broken wings or black undersides that obscured the medial line were not analized. The

Figure 1. Different degrees of acutenness of the forewing apex (straight, semiacute and acute) and of the elongation of vein M3 of the hind wings (absent, small, medium, long) in *Anaea ryphea* and *Anaea eurypyle* (modified from Comstock, 1961).



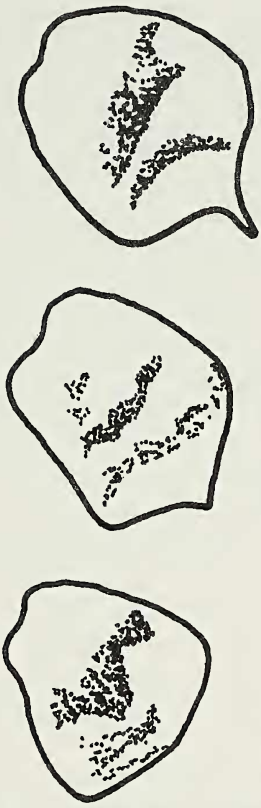


Figure 2. Different degrees of irregularity of the medial line on underside of the hind wings (straight, undefined, irregular) in *Anaea ryphea* and *Anaea eurypyle* (after figures in Comstock, 1961).

analysis was restricted to males because females of both taxa always have long tails, although they also vary in shape of the medial line and the forewings. I also did 40 genitalic dissections to look for the differences in the whole male apparatus of the two species, as shown by Comstock in drawings (1961, pp.164).

In order to assess the distribution of the two main distinguishing characters found after these preliminary analyses, I did a frequency distribution of them throughout both species combined as one, looking for a bimodal distribution that could support the existence of two species.

Results

Analysis of 475 specimens that were in good condition showed that:
1) Among a total of 165 males labelled *A. eurypyle*, 53% had long tails (>4mm), 47% had medium tails (2-4mm), and 0.6% (one) had small tails (<2mm). No individual was found without a tail. Among the 310 males labelled *A. ryphea*, 1% had long tails, 5% had medium tails, 71% had small tails, and 23% had no tail at all (rounded HW). Thus, if tail length alone were considered, specimens with long tails would separate as *A. eurypyle*, those with small to

non-existent tails would be *A. ryphea*, and those with medium length tails would be impossible to assign because a clear cutoff does not exist.
2) Of 165 males labelled *A. eurypyle*, 52% had a straight medial line on the underside of the HW; 48% had the line irregular to differing degrees (from slightly to moderate); and one individual had an extremely irregular line, but perhaps because it had a long tail, it was labelled *eurypyle*. Among the 310 males of *A. ryphea* analyzed, 91% had the medial line irregular to differing degrees (from moderate to extremely); 8% had it slightly irregular; and 2 had it straight, yet were labelled *ryphea*, perhaps because they had no tail. So, for this character, specimens with the medial line straight to moderately irregular would pertain to *eurypyle*, while those with the line moderately to extremely irregular would be assigned to *ryphea*, but again there is continuous variation. As well, individuals with a straight medial line didn't necessarily have the medium-long tails, nor did the ones with an irregular medial line always have a small tail or no tail, although a trend exists.
3) Most (61%) *A. eurypyle* males of both subspecies, *confusa* and *eurypyle*, had a, so-called, semi-acute forewing, while 11% had it acute, and 28% had it straight. According to Comstock (1961), semi-acute forewing is a seasonal variant in South American specimens (subspecies *eurypyle*) and a non-seasonal "random" variant in Central America (subspecies *confusa*). Apart from that, Comstock found straight forewings only in individuals from Central America, and considered it to be a subspecific character. Nevertheless, there are individuals from South America (mainly from Peru, various localities and dates) with straight-margined forewings. Analysis of individuals labeled *A. ryphea* showed a similar frequency distribution for this characteristic.
4) The frequency of black and white markings on the undersides of the wings was similar in both taxa. Comstock (1961) stated that they were "not uncommon" in *ryphea* and "occasional" in *eurypyle*, probably based on the limited series of specimens that he examined.
5) There is wide variation in genitalic characters, but the differences shown in the genitalic drawings of the two taxa in Comstock (1961) could not be found. A further genitalic study is needed to assess the whole variation within both species.

Discussion

I conclude that tail length and degree of irregularity of the medial line cannot be taken as definitely distinguishing *A. ryphea* and *A. eurypyle*. These characters vary continuously in both taxa, and no natural (or artificial) limits could be found to separate them. Also, many statements in the literature regarding species variation and separation are based on small or biased samples of specimens actually displaying random variation. Characters that would be expected to occur together in each taxon were found not necessarily to be correlated in every individual, as were supposedly seasonal characters. Thus, there are many individuals that cannot be included in any of the species because of mixed characters.

Continuous variation in morphological characters is not uncommon. A similar situation was found by Robbins (1991) in two species of *Mitoura* (Lycaenidae), where the second "species" was nothing more than an isolated example plucked from the morphological unimodal continuum of characters that define the original species. Burns (1984) has encountered examples of the same situation with skippers (Hesperiidae). A more refined quantification of the variable characters in *A. ryphea* and *A. eurypyle* will perhaps provide adequate data to elucidate the relationship between these two taxa, but a frequency distribution of the two main distinguishing characters (length of tail and degree of irregularity of medial line), when individuals of both species are combined, is clearly unimodal (Fig. 3). The fact that there is only one mode suggests that we could be really dealing with only one species. Specimens identified as *A. eurypyle* are individuals with wing patterns on one extreme of both distributions — long tail and regular mesial line. All the rest of both distributions, including the mode, would be considered *A. ryphea.* There is no significant difference between the proportions of individuals within each class of these distributions (Table 1), which shows that there is a tendency for individuals with long tails to have straight medial line; for those with medium tails to have undefined medial line; and for those with small or no tail to have irregular medial line. The two distinguishing characters vary together, in accordance, in the majority of the cases. That could also explain why *A. ryphea* is more common in all collections than *A. eurypyle.*

A very puzzling fact is that *Anaea eurypyle* is not widely recorded from Panama, although it is found throughout the whole range of *A. ryphea.* In fact, the only record for Panama is of five individuals collected near the border with Costa Rica, which were used by Comstock (1961) in his revision. These specimens are no longer in the AMNH, and I have not been able to trace them. Apparently, nobody since then has collected *A. eurypyle* in Panama (I went there again in 1994, for this specific purpose, but failed to find any), although records of *A. ryphea* are abundant.

Panamanian individuals reared by me in the laboratory during 1991/1992, and again in 1994, showed no great morphological differences among the females, but males varied greatly in color, pattern, and shape of the wings. The distal bars of the forewing dorsal surface ranged from bright blue to almost black (which is considered typical of *A. eurypyle*), and the basal area of the hindwings, near the thorax, varied from purple to orange. The elongation of vein M3 in the hind wings ranged from almost none to moderate. The ventral surface pattern was extremely variable, and one individual had a definitely straight medial line. This characteristic was found also in one specimen at the AMNH, and in several specimens at the USNM, all from Panama. Variability was also observed in adults from Campinas, Brazil, but the elongation of vein M3 did not occur there. Some of this variation has been cited (Comstock 1961) but has not been quantified in such a way as to permit assessment of its importance in the determination of the two species. *Anaea ryphea* and *A. eurypyle* could, in fact, represent one

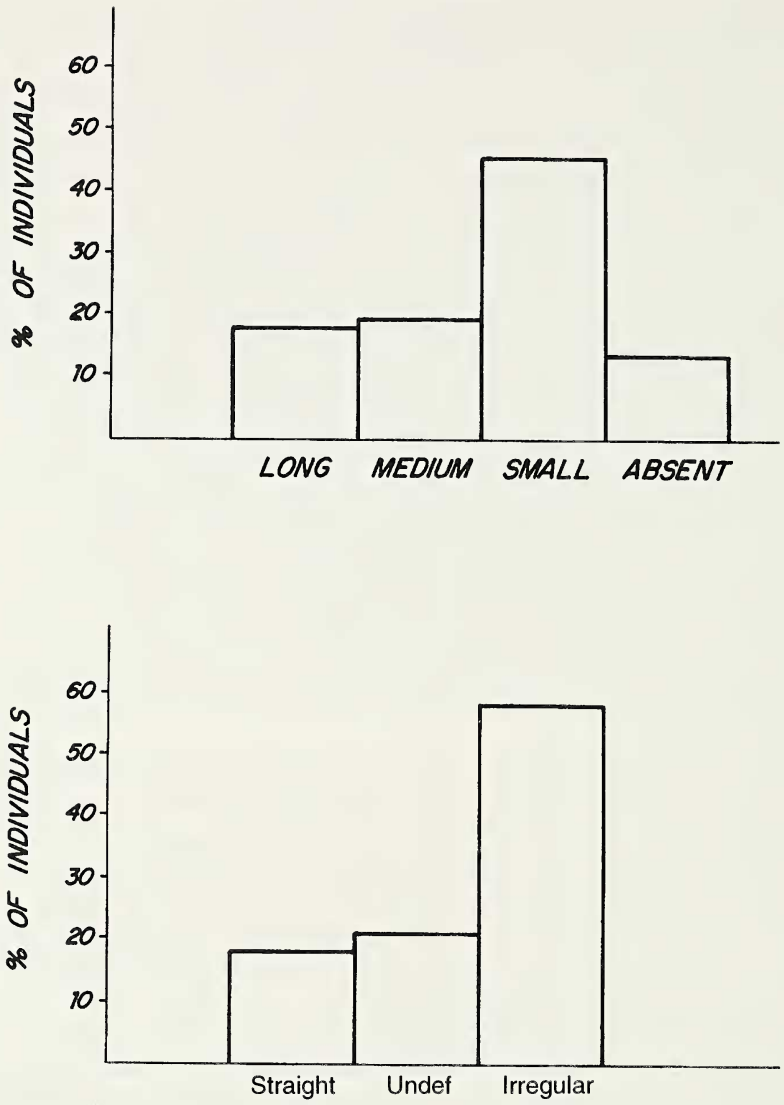


Figure 3. Frequency distributions of the two main distinguishing characters between *Anaea ryphea* and *Anaea eurypyle* — (a) length of tail and (b) degree of irregularity of medial line — with individuals of both species combined.

highly variable species, or if they are distinct biological species, they have considerable character overlap (Mayr and Ashlock 1991). Perhaps molecular studies will shed more light on the situation.

Temperature and relative humidity during development can influence adult morphological characters in Lepidoptera. It is also known that widely distributed species tend to develop locally differentiated populations and to show generally high levels of variability (Ehrlich and Raven 1969; Endler 1973, 1977), usually related to ecological and biogeographical factors.

Table 1. Test for proportions of individuals within each class of the frequency distribution for the two distinguishing characters between *Anaea ryphea* and *Anaea eurypyle*.

Tail length	Medial line	z	p
long - 19%	straight - 19%	0.000	0.500
medium - 20%	undefined - 22%	-0.757	0.225
small/absent - 61%	irregular - 59%	-0.629	0.266

Variability can then be a consequence of genetic and environmental factors combined. Given that *A. ryphea* has an extensive geographical distribution, very likely it has developed local patterns of differentiation.

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Literature Cited

Burns, J.M. 1984. Evolutionary Differentiation: Differentiating Gold-banded Skippers — *Autochton cellus* and More (Lepidoptera: Hesperidae: Pyrginae). Smithsonian Contributions to Zoology 405. Smithsonian Institution Press, Washington, DC. 38pp.

—— . 1992. Genitalic characterization, enlargement, and reassociation of the Neotropical Hesperiinae genus *Halotus* (Hesperiidae). J. Lepid. Soc. 46(3): 182-194.

Caldas, A. 1991. A population of *Anaea ryphea* (Nymphalidae) and its larval foodplant at Campinas, Brazil. J. Lepid. Soc. 45: 68.

——. 1994. Biology of *Anaea ryphea* at Campinas, Brazil. J. Lepid. Soc. 48(3): 248-257.

——. 1995a. Mortality of *Anaea ryphea* (Nymphalidae) immatures in Panama. J. Res. Lepid. 31(3-4): 195-204.

—— . 1995b. Population ecology of *Anaea ryphea* (Nymphalidae): immatures at Campinas, Brazil. J. Lepid. Soc. 49(2): 234-245.

Collins, M.M. 1991. Speciation: a review of concepts and studies with special reference to the Lepidoptera. J. Res. Lepid. 30: 45-81.

Comstock, W.P. 1961. Butterflies of the American tropics. The genus *Anaea*, Lepidoptera, Nymphalidae. American Museum of Natural History, New York. 214 pp.

Descimon, H. 1986. The evolution of coloration in Neotropical Charaxidae: adaptive strategies and cladogenesis (Lepidoptera, Rhopalocera). Bull. Soc. Zool. Fr. 111: 261-296.

DeVries, P.J. 1987. The butterflies of Costa Rica and their natural history. Princeton University Press, New Jersey. 327 pp.

Ehrlich, P.R. & P.H. Raven. 1969. Differentiation of populations. Science 165: 1228-1232.

Endler, J.A. 1973. Gene flow and population differentiation. Science 179: 243-250.

——. 1977. Geographic variation, speciation, and clines. Princeton Univ. Press, New Jersey.

Mayr, E. & P.D. Ashlock. 1991. Principles of Systematic Zoology. McGraw Hill, New York. 2.ed. 475p.

Muyshondt, A. 1974. Notes on the life cycle and natural history of butterflies of El Salvador. VI. *Anaea* (*Memphis*) *eurypyle confusa* (Nymphalidae). J. Lepid. Soc. 28: 306-314.

Owen, D.F. 1971. Tropical butterflies. Clarendon, Oxford.

Robbins, R.K. 1990. The *Mitoura spinetorum* complex in New Mexico and the validity of *M. millerorum* (Lycaenidae: Theclinae). J. Lepid. Soc. 44(2): 63-76.

Rydon, A.H.B. 1971. The systematics of the Charaxidae (Lepidoptera: Nymphalidae). Entomologist's Rec. J. Var. 83: 339-341.

Vane-Wright, R.I. & P.R. Ackery (eds). 1984. The biology of butterflies. Academic Press, London.